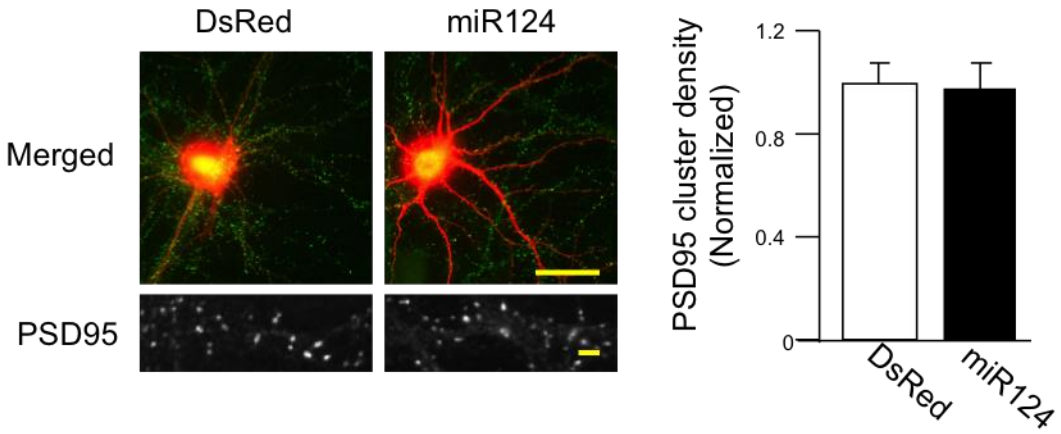
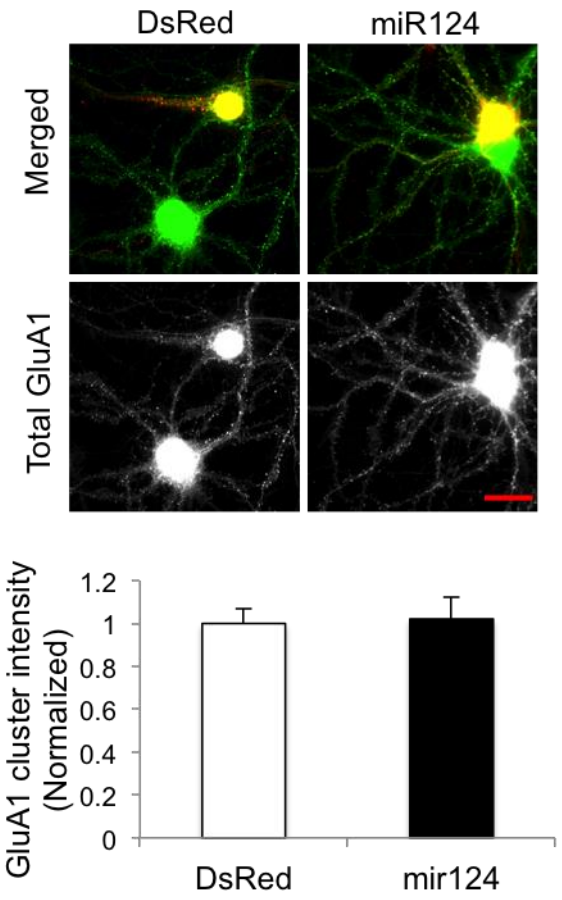
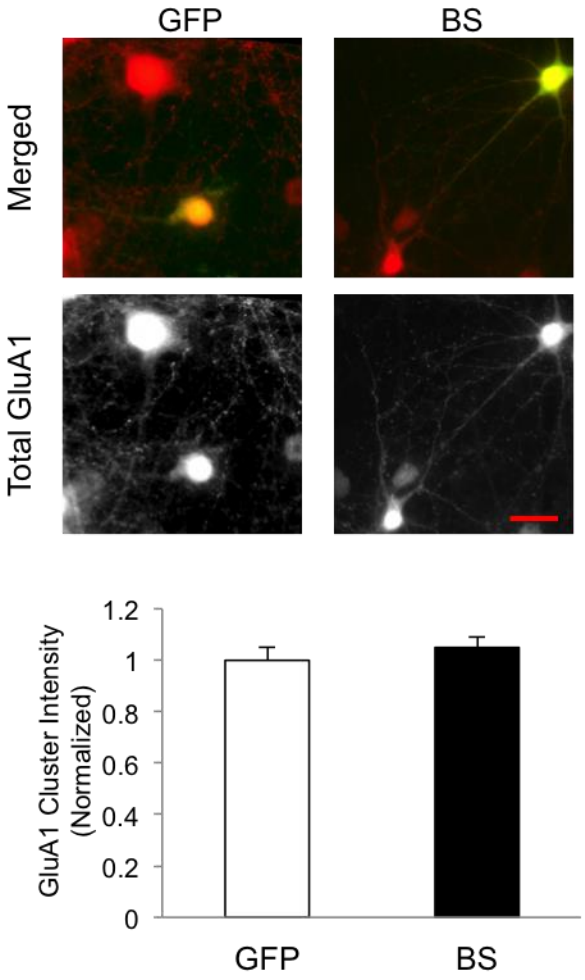


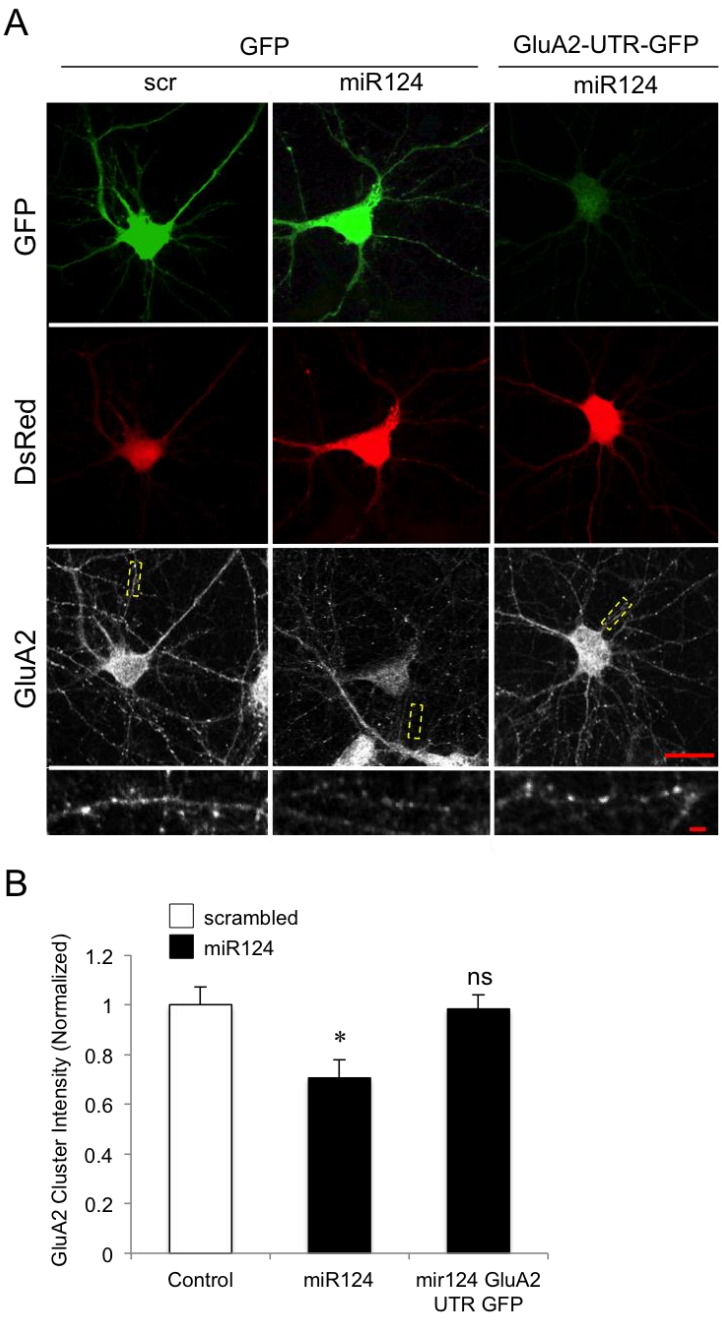
Supplementary Figure 1. miR124 does not change neuron morphology and synaptic density. Hippocampal neurons were transfected with miR124 (containing DsRed) or DsRed as a control. 2 d after transfection, neurons were immunostained for PSD95 (green). Expression of miR124 did not cause marked changes in neurite structure and density of PSD95 clusters. Bar graph represents Mean ± SE, n = 16 cells, $p>0.05$, t test. Scale bars = 10 μm (full images), 3 μm (dendrites).



Supplementary Figure 2. miR124 does not affect GluA1 expression. Hippocampal neurons were transfected with miR124 (containing DsRed) or DsRed as a control. 2 d after transfection, neurons were immunostained for GluA1 (green) under permeant conditions. No changes in GluA1 cluster intensity were found in cells expressing miR124. Bar graphs represent Mean ± SE. n = 18 cells, t test. Image scale bar = 10 μm.

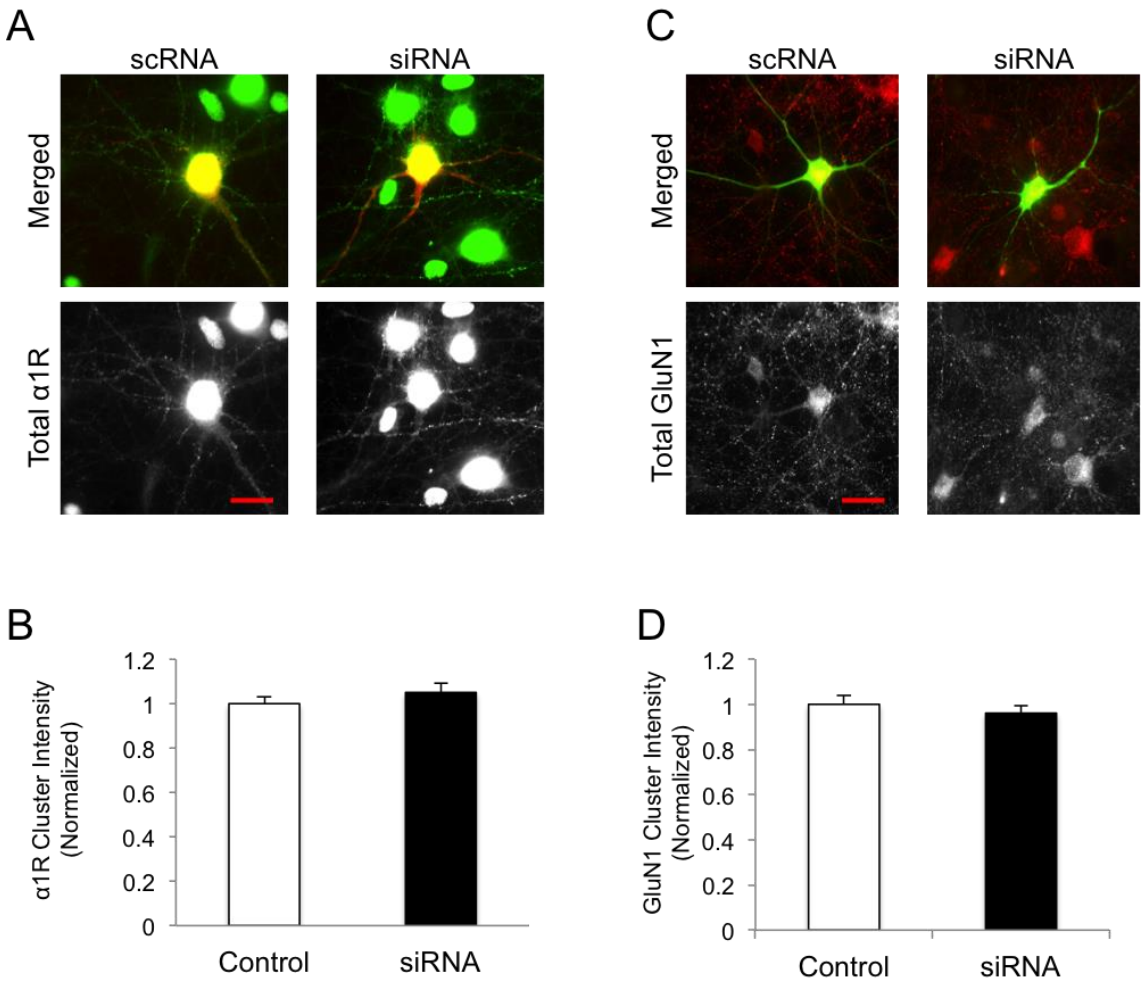


Supplementary Figure 3. miR124 BS does not affect total GluA1 expression. Cultured hippocampal neurons were transfected with a miR124-neutralizing sponge BS (containing GFP) at DIV12 and total GluA1 was immunostained 2 d later. GFP was transfected as a control. In cells expressing miR124 BS, the total GluA1 puncta intensity was not affected. Bar graph represents Mean ± SE, n = 10 cells, $p>0.05$, t test. Image scale bar = 10 μm.

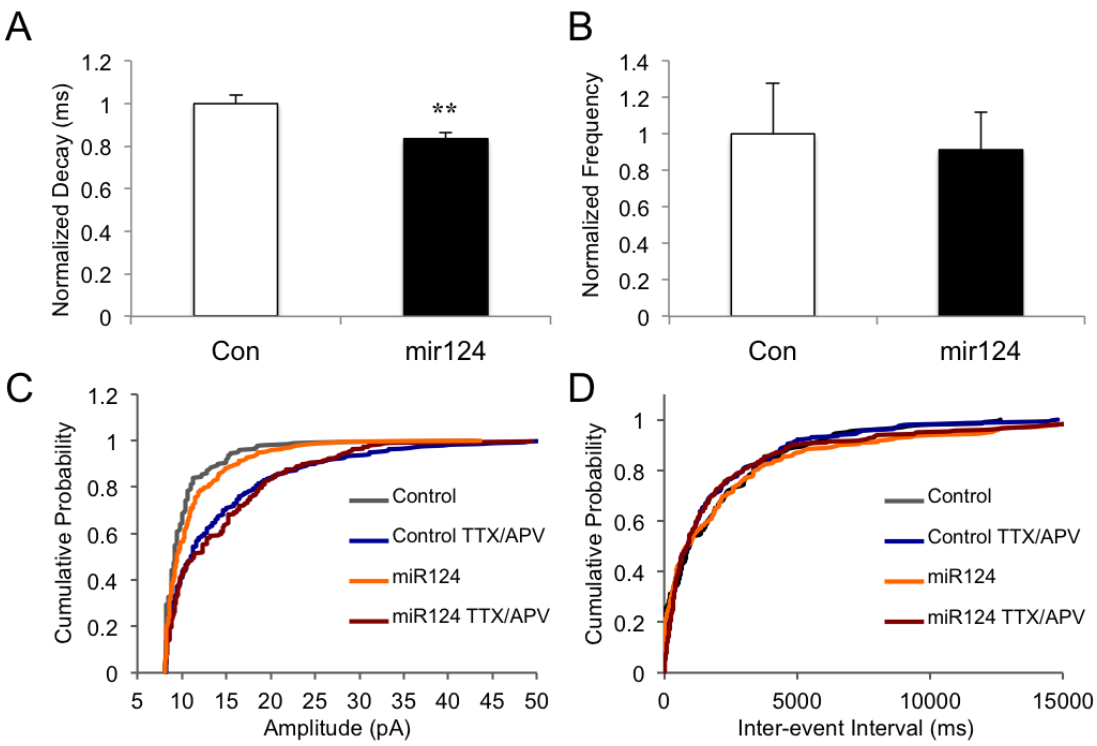


Supplementary Figure 4. GluA2-UTR-GFP blocks the miR124-mediated decrease in GluA2 expression. (A) DIV12 cultured hippocampal neurons were co-transfected with miR124 or a scrambled control (both contain DsRed) with GFP, or co-transfected with GluA2-UTR-GFP with miR124. Total GluA2 was immunostained 2 d later under permeant conditions.

(B) In cells expressing miR124, GluA2 levels were decreased compared to the scrambled control. However, miR124 did not change GluA2 in neurons expressing GluA2-UTR-GFP. Bar graph represents Mean ± SE, n = 9 - 10 cells, * = $p<0.05$, ns = $p>0.05$, t test. Image scale bars = 10 μm (full images), 3 μm (dendrites).

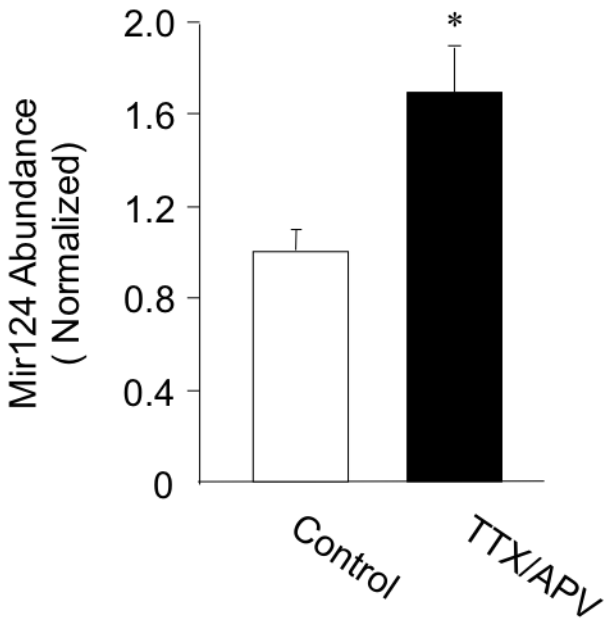


Supplementary Figure 5. Suppression of miR124 does not affect GABAR and NMDAR expression. Hippocampal neurons were transfected with siRNA against miR124, together with DsRed. Scrambled siRNA (scRNA) was used as a control. GABA receptor subunit α1 (α1R) or NMDA receptor subunit GluN1 were immunostained 2 d after transfection. Inhibition of miR124 did not affect the expression of α1R (A and B; n = 10 cells, $p>0.05$, t test) or GluN1 (C and D; n = 11 cells, $p>0.05$, t test). Image scale bars = 10 μm.

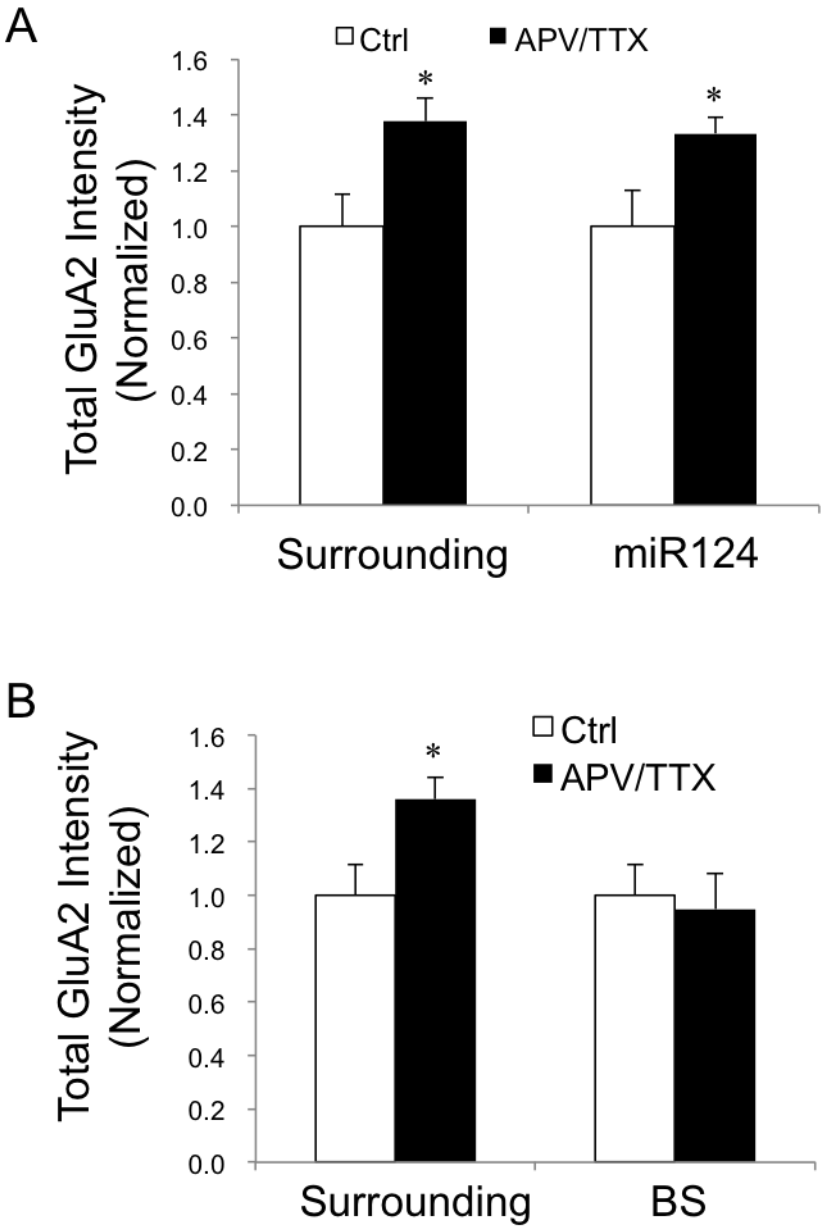


Supplementary Figure 6. miR124-transfected cells show decreased mEPSC decay time and typical multiplicative homeostatic scaling. (A) miR124 expressing cells showed a decreased in average mEPSC decay time. Bar graph represents Mean ± SE, n = 6 cells, ** = $p<0.01$, t test. (B) miR124 expression did not change mEPSC frequency. Bar graph represents Mean ± SE, n = 6 cells, $p>0.05$, t test.

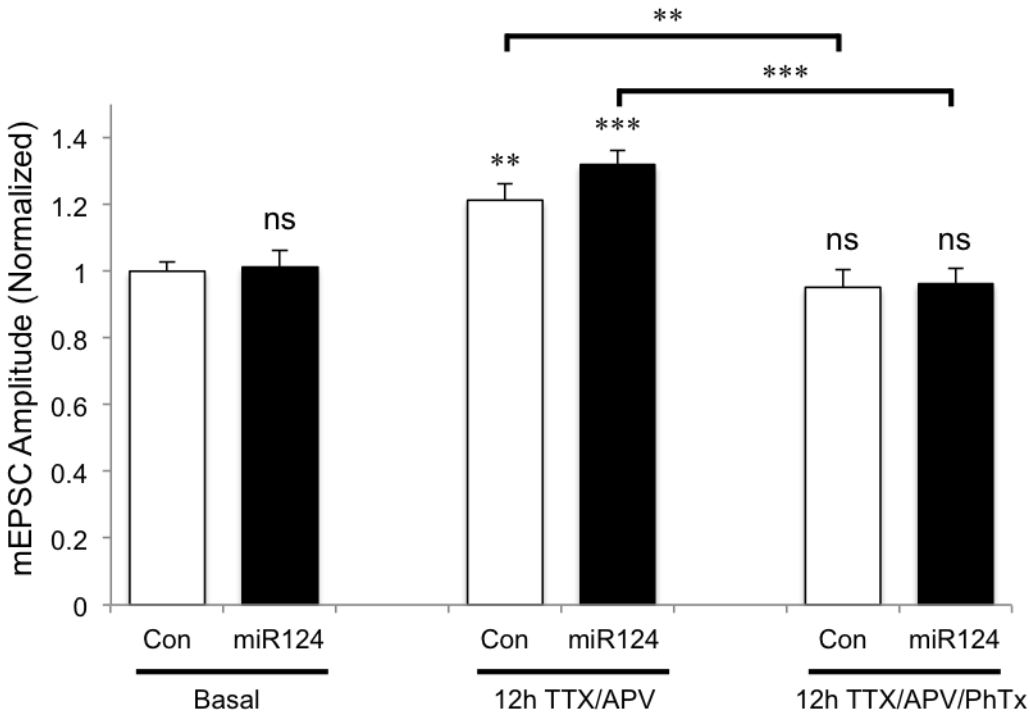
(C and D) Cumulative probability plots of amplitude (C) and inter-event interval (D) show typical multiplicative scaling by TTX/APV incubation in miR124-transfected cells.



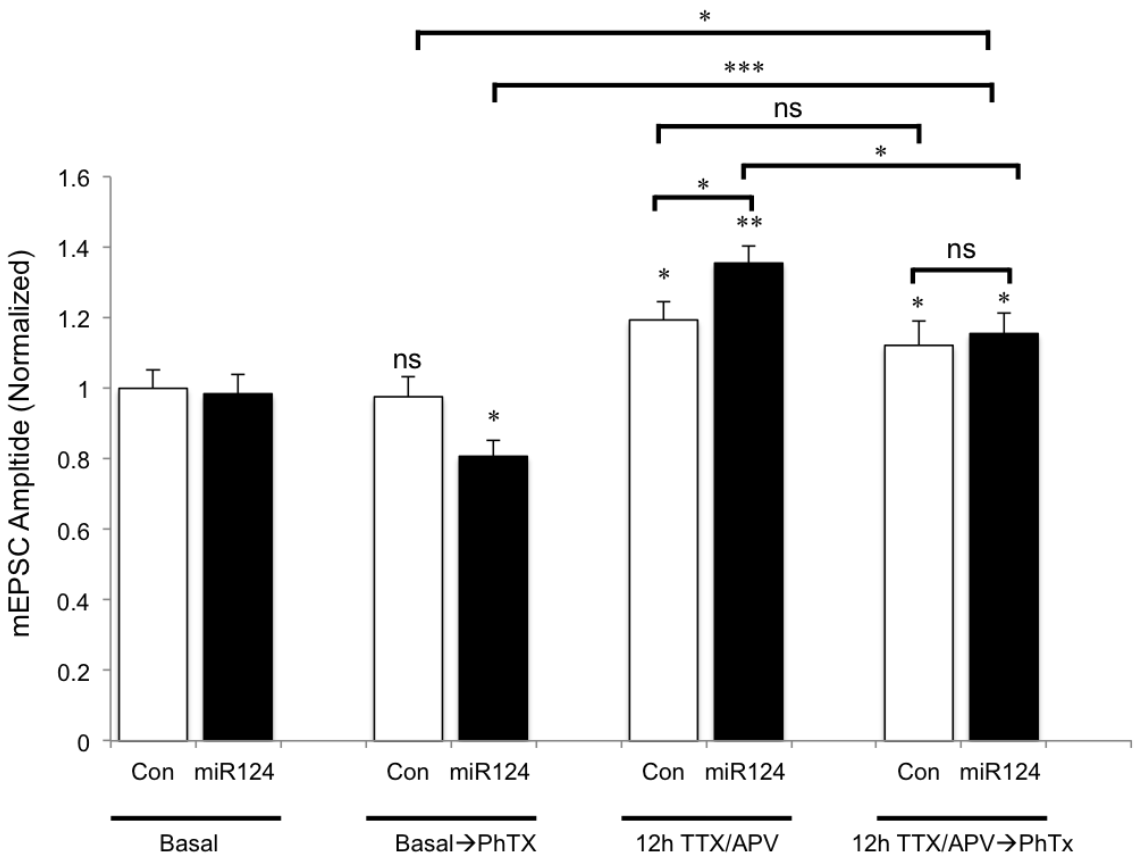
Supplementary Figure 7. Neuronal inactivity causes an increase in mir124 expression. Hippocampal neurons were incubated with TTX/APV for 15 hr. The amount of mir124 was measured by qPCR. Neuronal inhibition led to an increase in mir124. ($n = 3$, $p<0.05$, t test).



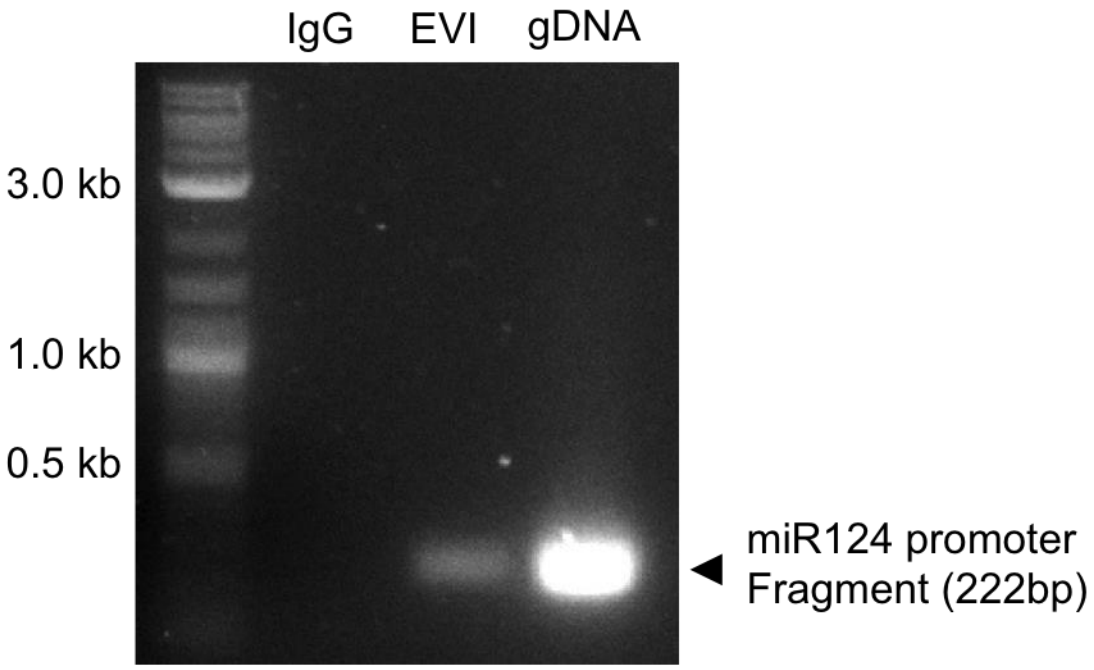
Supplementary Figure 8. GluA2 is increased during HSP. Neurons were transfected with mir124 or BS for 1 d, and then incubated with TTX/APV for 15 hr. Immunostaining showed a significant increase in GluA2 puncta intensity in both the non-transfected surrounding cells and the transfected cells (A). The homeostatic increase in GluA2 was blocked by BS (B).



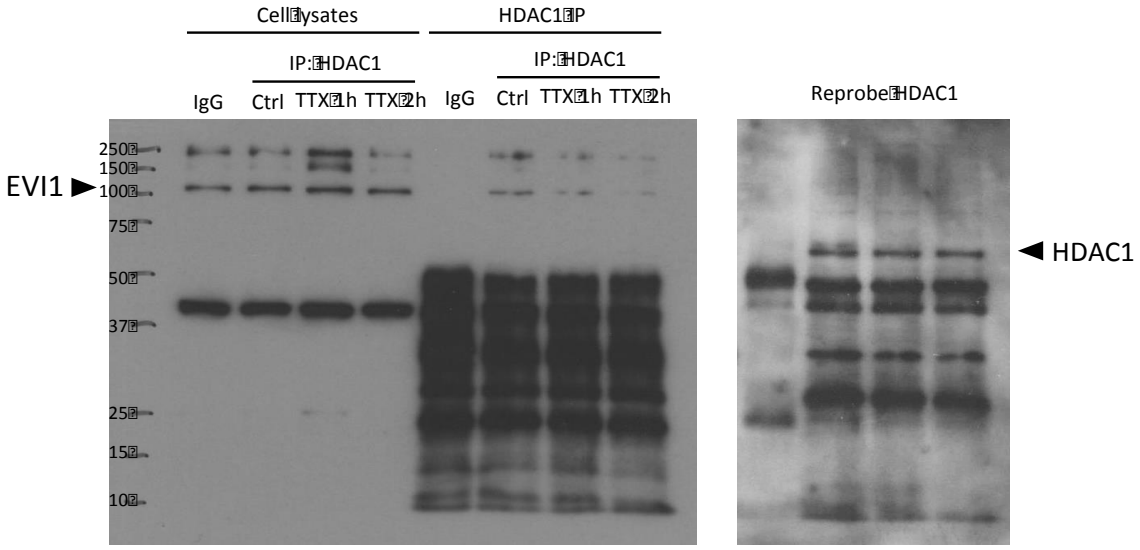
Supplementary Figure 9. CP-AMPARs are required for HSP. Hippocampal neurons were transfected with mir124 for 1 d, and then incubated with TTX/APV, with or without PhTx, for 12 hr. Application of PhTx abolished HSP expression shown by mEPSC recordings. (n = 5-8 cells, * = $p<0.05$, ** = $p < 0.01$, *** = $p<0.001$, ns = $p>0.05$, t test).



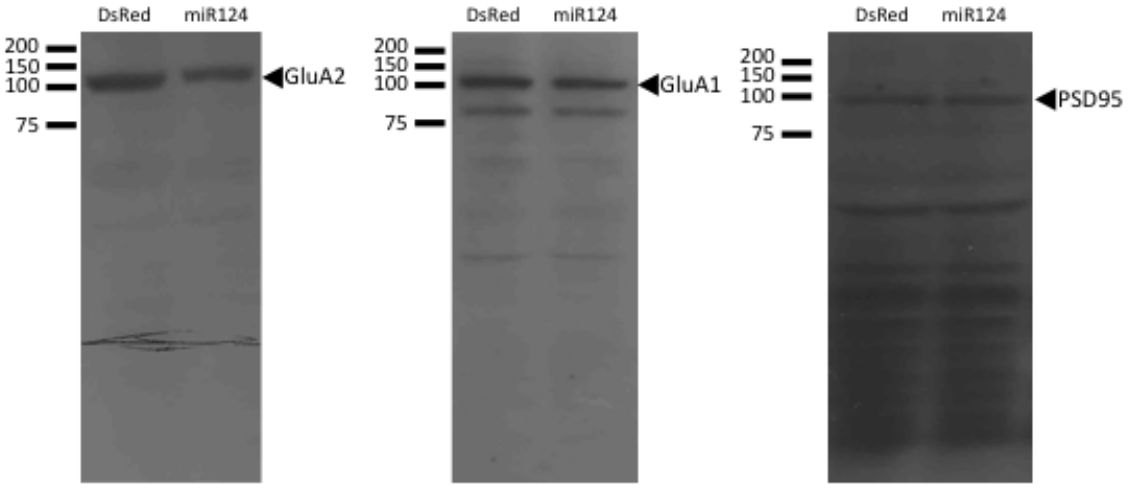
Supplementary Figure 10. Contribution of GluA2-containing AMPARs in HSP. Mir124-transfected neurons were incubated with TTX/APV for 12 hrs to induce HSP. Cells were then recorded in the absence and presence of PhTx. PhTx caused a decrease in mEPSC amplitude in mir124-transfected cells under control conditions (2nd pair of bar graphs). In TTX/APV treated cells, PhTx caused a partial decrease in mEPSC amplitude in mir124 transfected cells, but not in the control cells (3rd and 4th pairs of bar graphs) (n = 6-9 cells, * = $p<0.05$, ** = $p<0.01$, *** = $p<0.001$, ns = $p>0.05$, t test).



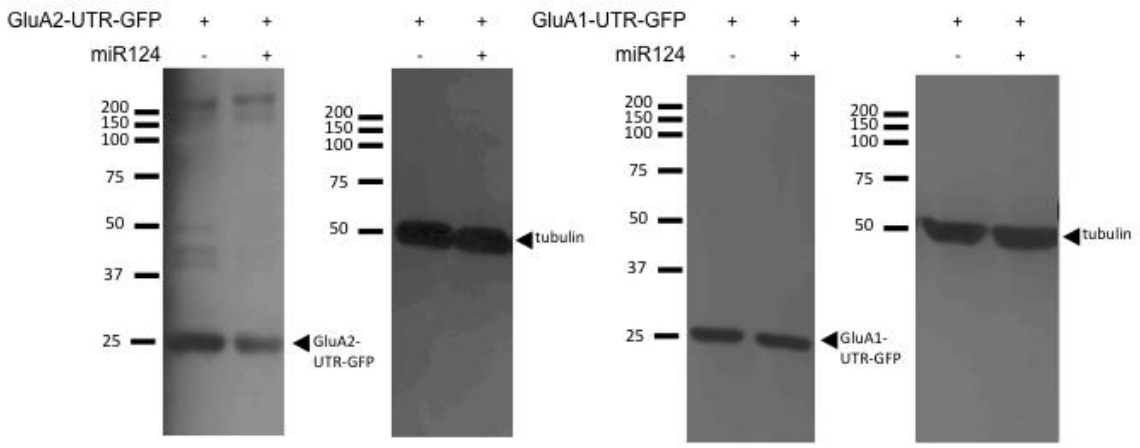
Supplementary Figure 11. Association of EVI1 with mir124 promoter. A ChIP assay shows positive detection of the miR124 promoter sequence in EVI1 immunoprecipitates from cortical neurons. Genomic DNA (gDNA) was used as control.



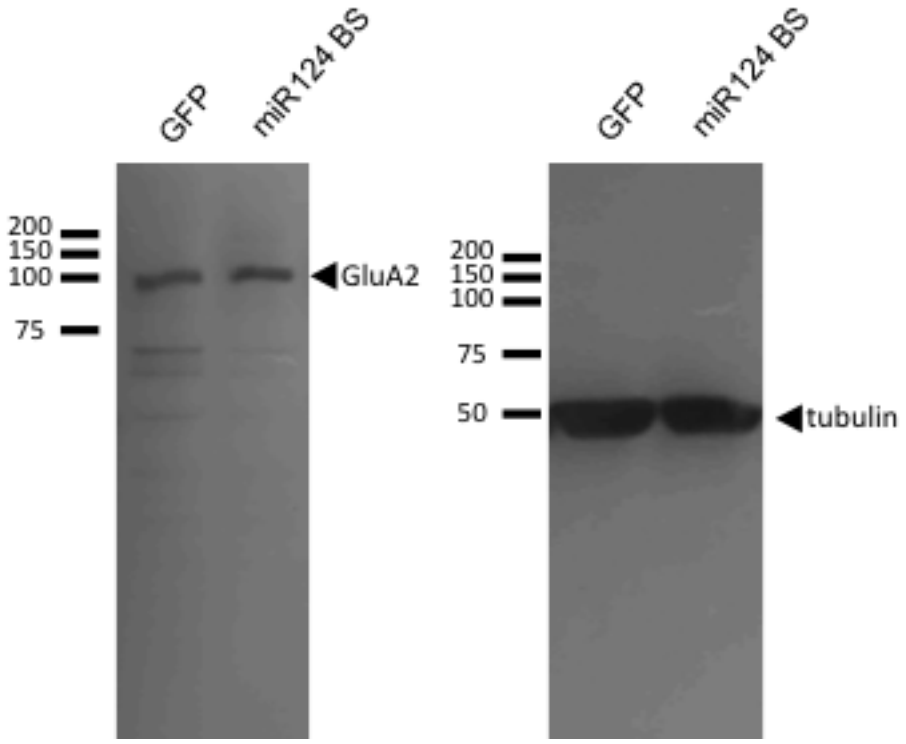
Supplementary Figure 12. Co-IP of EVI1 and HDAC1. Neurons were incubated with TTX/APV for 1 or 2 hr. Using the cell lysates, immunoprecipitates of anti-HDAC1 antibodies were probed for EVI1. HDAC1-EVI1 association was reduced by neuronal inhibition.



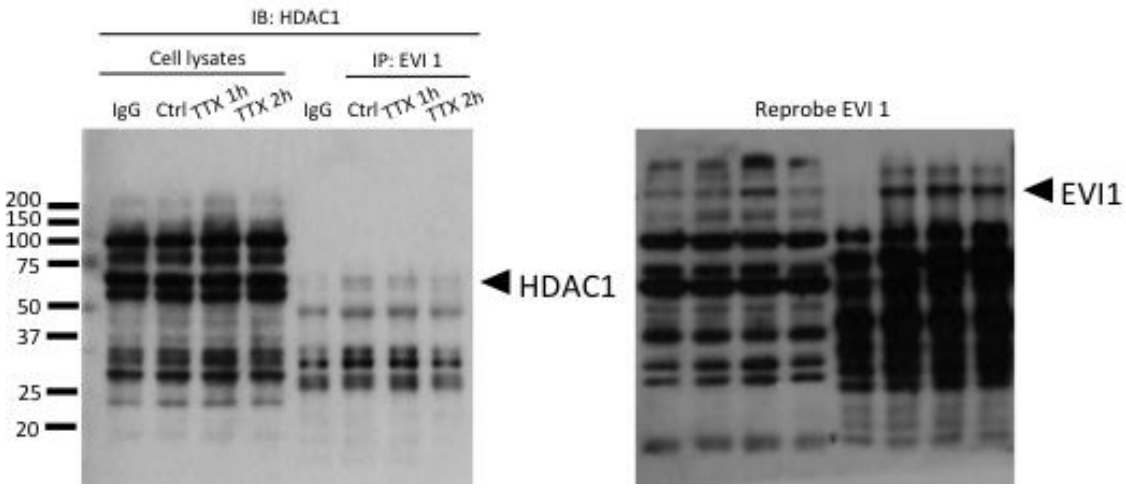
Supplemental Figure 13. Full western blots for Figure 1E.



Supplemental Figure 14. Full western blots for Figure 2B.



Supplemental Figure 15. Full western blots for Figure 3D.



Supplemental Figure 16. Full western blots for Figure 7G.

Treatment	Amplitude (pA)	SE
Con Basal	12.77	0.37
miR124 Basal	12.95	0.62
Con TTX/APV	15.48	0.65
miR124 TTX/APV	17.83	0.55
Con TTX/APV/PhTx	12.15	0.66
miR124 TTX/APV/PhTx	12.30	0.60

Table 1. mEPSC amplitudes from Supplementary Figure 9.

Treatment	Amplitude (pA)	SE
Con Basal	13.82	0.48
miR124 Basal	13.84	0.78
Con Basal → PhTx	13.74	0.79
miR124 Basal → PhTx	11.37	0.65
Con TTX/APV	16.80	0.71
miR124 TTX/APV	19.08	0.68
Con TTX/APV → PhTx	15.77	0.59
miR124 TTX/APV → PhtX	16.42	0.57

Table 2. mEPSC amplitudes from Supplementary Figure 10.